

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

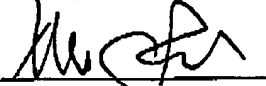
Page 2 [Amendment Under 37 C.F.R. §1.115

(In Response To The September 8, 2006 Office Action)]

- October 19, 2006

RECEIVED  
CENTRAL FAX CENTER

OCT 19 2006

CERTIFICATE OF TRANSMISSION	
I hereby certify that this paper (Amendment Under 37 C.F.R. §1.115) (along with any documents referred to as attached or enclosed) is being facsimile transmitted to the United States Patent and Trademarks (Fax No. 571-273-8300) on October 19, 2006.	
10-19-06 Date	 Ronald C. Fedus Reg. No. 32,567

Amendments to the Claims are reflected in the listing of claims which begins on page 3 of this paper.

Remarks begin on page 12 of this paper.

**Amendments to the Claims:**

The listing of the claims below will replace all prior versions and listing of claims in this application.

Enz-61(D11)

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 3 [Amendment Under 37 C.F.R. §1.115

(In Response To The September 8, 2006 Office Action)]

- October 19, 2006

RECEIVED  
CENTRAL FAX CENTER

OCT 19 2006

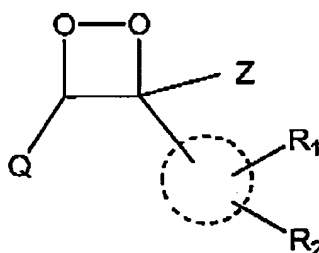
**Listing of Claims:**

Claims 1-286 (Canceled).

287. (Previously Presented) A process for detecting the presence or quantity of enzymatic activity of interest in a sample, said process comprising the steps of:

(a) providing:

- (i) said sample suspected of containing enzymatic activity;
- (ii) a chemiluminescent reagent having the structure:



wherein Q comprises a cycloalkyl or polycycloalkyl group attached covalently to the 4-membered ring portion of said dioxetane above directly or indirectly through a linkage group; wherein Z comprises hydrogen, alkyl, aryl, aralkyl, alkaryl, heteroalkyl, heteroaryl, cycloalkyl or cycloheteroalkyl; and wherein R<sub>1</sub> and R<sub>2</sub> comprise chemical moieties attached to different sites of a cyclic ring attached to said dioxetane, and wherein R<sub>1</sub> is enzymatically converted into R<sub>1</sub>\* which comprises a chemical reactive group G<sub>1</sub>, and wherein R<sub>2</sub> is attached to said cyclic ring through an oxygen atom and comprises a chemical reactive group G<sub>2</sub> which reacts with said G<sub>1</sub> to convert said dioxetane to an unstable light-emitting dioxetane form.

Enz-61(D11)

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 4 [Amendment Under 37 C.F.R. §1.115

(In Response To The September 8, 2006 Office Action)]

– October 19, 2006

- (ii) reagents and buffers for carrying out chemiluminescent reactions;
- (b) forming a mixture of:
- (1) (i), (ii) and (iii); or
- (2) (ii) and (iii) and contacting said mixture of (ii) and (iii) with (i);
- (c) enzymatically converting the chemiluminescent reagent of (a)(ii) into an unstable light-emitting dioxetane form; and
- (d) measuring the quantity of light generated by said enzymatic conversion in step (c).

288. (Previously Presented) The process of claim 287, wherein in said providing step (a) Q in said chemiluminescent reagent (ii) comprises an adamantyl group.

289. (Previously Presented) The process of claim 287, wherein in said providing step (a) R<sub>2</sub> in said chemiluminescent reagent (ii) comprises a substituted or unsubstituted aliphatic group or an unsubstituted aromatic group.

290. (Currently Amended) The process of claim 289, wherein said substituted aliphatic group comprises halogen, ~~nitrates~~, or sulfonates ~~or nitrites~~.

291. (Currently Amended) The process of claim 287, wherein said enzymatic converting step (c) is carried out by a substrate comprising ~~amides, esters, phosphates, carboxylic acids, fatty acids~~, glucose, xylose, fucose, ~~or~~ amino acids, or esters of phosphates, carboxylic acids or fatty acids.

Enz-61(D11)

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 5 [Amendment Under 37 C.F.R. §1.115

(In Response To The September 8, 2006 Office Action)]

- October 19, 2006

292. (Currently Amended) The process of claim 287, wherein said enzymatic activity of interest comprises an amidase, ~~an esterase, an acetylcholinesterase, an acid phosphatase, an alkaline phosphatase, a decarboxylase, a lipase, a glucosidase, a xylosidase, a fucosidase,~~ a trypsin or a chymotrypsin.

293. (Previously Presented) The process of claim 287, further comprising the step of forming an intermediate five- or six-membered ring comprising a linkage between said G<sub>1</sub> and G<sub>2</sub> in said chemiluminescent reagent (ii).

294. (Previously Presented) The process of claim 287, wherein any of said steps (a) through (d) are carried out in liquid phase or mixed phase.

295. (Previously Presented) The process of claim 287, wherein said enzymatic activity of interest is dependent upon the presence or quantity of another compound.

296. (Previously Presented) The process of claim 295, wherein said another compound comprises an RNA or DNA probe.

Enz-61(D11)

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 6 [Amendment Under 37 C.F.R. §1.115

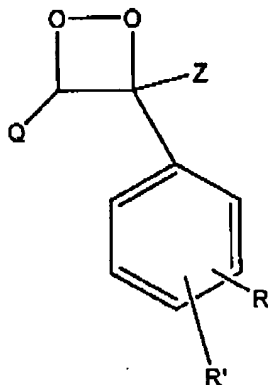
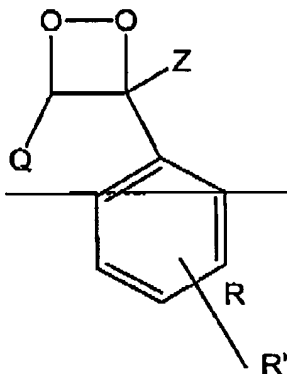
(In Response To The September 8, 2006 Office Action)]

- October 19, 2006

297. (Currently Amended) A process for detecting the presence or quantity of enzymatic activity of interest in a sample, said process comprising the steps of:

(a) providing:

- (i) said sample suspected of containing enzymatic activity;
- (ii) a chemiluminescent reagent having the structure:



wherein Q comprises a cycloalkyl or polycycloalkyl group attached covalently to the 4-membered ring portion of said dioxetane above directly or indirectly through a linkage group; wherein Z comprises hydrogen, alkyl, aryl, aralkyl, alkaryl,

Enz-61(D11)

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 7 [Amendment Under 37 C.F.R. §1.115

(In Response To The September 8, 2006 Office Action)]

- October 19, 2006

heteroalkyl, heteroaryl, cycloalkyl or cycloheteroalkyl; and wherein R comprises a chemical linker having a reactive site attached to the aromatic ring in said structure; and wherein R' comprises a substrate for an non-cleaving enzymatic process, wherein the product of said enzymatic process leads to further chemical rearrangements that generate an unstable light emitting dioxetane form.

(ii) reagents and buffers for carrying out chemiluminescent reactions;

(b) forming a mixture of:

(1) (i), (ii) and (iii); or

(2) (ii) and (iii) and contacting said mixture of (ii) and (iii) with (i);

(c) enzymatically converting the chemiluminescent reagent of (a)(ii) into an unstable light-emitting dioxetane form; and

(d) measuring the quantity of light generated by said enzymatic conversion in step (c).

298. (Previously Presented) The process of claim 297, wherein in said providing step (a) Q in said chemiluminescent reagent (ii) comprises an adamantyl group.

299. (Previously Presented) The process of claim 297, wherein in said providing step (a) R in the chemiluminescent reagent (ii) comprises a substituted or unsubstituted aliphatic group or an unsubstituted aromatic group.

300. (Currently Amended) The process of claim 299, wherein said substituted aliphatic group comprises halogen, ~~nitrate~~, or sulfonate ~~or nitrite~~.

Enz-61(D11)

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 8 [Amendment Under 37 C.F.R. §1.115

(In Response To The September 8, 2006 Office Action)]

- October 19, 2006

301. (Previously Presented) The process of claim 297, wherein said providing step (a) R in the chemiluminescent reagent (ii) comprises a reactive site comprising an oxygen, a nitrogen or a sulfur atom.

302. (Previously Presented) The process of claim 297, wherein said step of enzymatically converting (c) is carried out by an enzyme comprising an oxidase or reductase.

303. (Previously Presented) The process of claim 297, wherein any of said steps (a) through (d) are carried out in liquid phase or mixed phase.

304. (Previously Presented) The process of claim 297, wherein said enzymatic activity of interest is dependent upon the presence or quantity of another compound.

305. (Previously Presented) The process of claim 304, wherein said another compound comprises an RNA or DNA probe.

306. (New) A process for detecting the presence or quantity of enzymatic activity of interest in a sample, said enzymatic activity being dependent upon the presence or quantity of another compound, said process comprising the steps of:

(a) providing:

(i) said sample suspected of containing enzymatic activity that is dependent upon the presence or quantity of said another compound;

(ii) a chemiluminescent reagent having the structure:

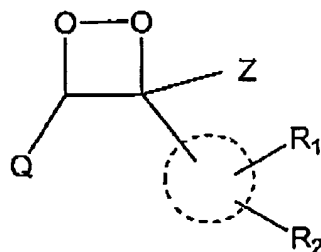
Enz-61(D11)

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 9 [Amendment Under 37 C.F.R. §1.115

(In Response To The September 8, 2006 Office Action)]

- October 19, 2006



wherein Q comprises a cycloalkyl or polycycloalkyl group attached covalently to the 4-membered ring portion of said dioxetane above directly or indirectly through a linkage group; wherein Z comprises hydrogen, alkyl, aryl, aralkyl, alkaryl, heteroalkyl, heteroaryl, cycloalkyl or cycloheteroalkyl; and wherein R<sub>1</sub> and R<sub>2</sub> comprise chemical moieties attached to different sites of a cyclic ring attached to said dioxetane, and wherein R<sub>1</sub> is enzymatically converted into R<sub>1</sub>\* which comprises a chemical reactive group G<sub>1</sub>, and wherein R<sub>2</sub> is attached to said cyclic ring through an oxygen atom and comprises a chemical reactive group G<sub>2</sub> which reacts with said G<sub>1</sub> to convert said dioxetane to an unstable light-emitting dioxetane form.

(ii) reagents and buffers for carrying out chemiluminescent reactions;

(b) forming a mixture of:

(1) (i), (ii) and (iii); or

(2) (ii) and (iii) and contacting said mixture of (ii) and (iii) with (i);

(c) enzymatically converting the chemiluminescent reagent of (a)(ii) into an unstable light-emitting dioxetane form; and

(d) measuring the quantity of light generated by said enzymatic conversion in step (c).

Enz-61(D11)



Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 10 [Amendment Under 37 C.F.R. §1.115

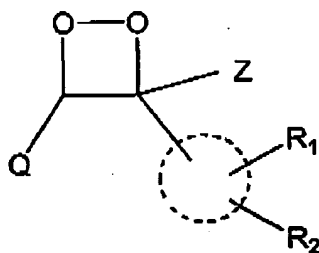
(In Response To The September 8, 2006 Office Action)]

- October 19, 2006

307. (New) A process for detecting the presence or quantity of enzymatic activity of interest in a sample, said enzymatic activity being dependent upon the presence or quantity of an RNA or DNA probe, said process comprising the steps of:

(a) providing:

- (i) said sample suspected of containing enzymatic activity that is dependent upon the presence or quantity of said RNA or DNA probe;
- (ii) a chemiluminescent reagent having the structure:



wherein Q comprises a cycloalkyl or polycycloalkyl group attached covalently to the 4-membered ring portion of said dioxetane above directly or indirectly through a linkage group; wherein Z comprises hydrogen, alkyl, aryl, aralkyl, alkaryl, heteroalkyl, heteroaryl, cycloalkyl or cycloheteroalkyl; and wherein R<sub>1</sub> and R<sub>2</sub> comprise chemical moieties attached to different sites of a cyclic ring attached to said dioxetane, and wherein R<sub>1</sub> is enzymatically converted into R<sub>1</sub><sup>\*</sup> which comprises a chemical reactive group G<sub>1</sub>, and wherein R<sub>2</sub> is attached to said cyclic ring through an oxygen atom and comprises a chemical reactive group G<sub>2</sub> which reacts with said G<sub>1</sub> to convert said dioxetane to an unstable light-emitting dioxetane form.

- (ii) reagents and buffers for carrying out chemiluminescent reactions;

Enz-61(D11)

Stravrianopoulos et al.; Serial No.: 10/764,389; Filed: January 23, 2004

Page 11 [Amendment Under 37 C.F.R. §1.115

(In Response To The September 8, 2006 Office Action)]

- October 19, 2006

- (b) forming a mixture of:
  - (1) (i), (ii) and (iii); or
  - (2) (ii) and (iii) and contacting said mixture of (ii) and (iii) with (i);
- (c) enzymatically converting the chemiluminescent reagent of (a)(ii) into an unstable light-emitting dioxetane form; and
- (d) measuring the quantity of light generated by said enzymatic conversion in step (c).

\* \* \* \* \*

Enz-61(D11)